AMENDMENTS TO THE CLAIMS

1. (Currently Amended) Method of preparing a pharmaceutical composition of a purified biologically active protein, comprising the steps of

adding to a solution of the purified protein a non-ionic detergent comprising polysorbate in an amount of 0.05 to 1 g/l;

subjecting the solution containing the non-ionic detergent to filtration on a virus removal filter with a pore size of 10 to 40 nm; and

recovering the filtrate.

2. (Original) The method according to claim 1, wherein the ~~non-ionic detergent~~ polysorbate is selected from the group consisting of polyoxyethylene sorbitan mono-oleate, and polyoxyethylene sorbitan monolaurate ~~and polyoxyethylene lauryl ether~~.

3. (Original) The method according to claim 1, ~~2,~~ wherein the non-ionic detergent comprises polyoxyethylene sorbitan mono-oleate ~~(polysorbate 80), which is added in an amount exceeding the critical micellar concentration~~.

4. (Cancelled)

5. (Previously Presented) The method according to any of claims 1 to 3, ~~to 4,~~ wherein the pharmaceutical composition comprises a solution of purified α-interferon.

6. (Previously Presented) The method according to claim 5, wherein the α-interferon solution before virus filtration, has an activity in the range of 3 to 50 mill. IU/ml.

7. (Previously Presented) The method according to claim 5, wherein the pharmaceutical composition comprises an α-interferon solution containing at least one α-interferon subtype selected from the group consisting of α1, α2, α4, α7, α8, α10, α14, α17 and α21.

8. (Previously Presented) The method according to claim 1, comprising preparing a pharmaceutical composition comprising purified leukocyte or lymphoblastoid α-interferon essentially in the absence of α-interferon polymers and albumin-interferon complexes.

9. (Previously Presented) The method according to claim 1, further comprising prefiltering said solution obtained in step (1) with a 0.04-0.2 μm filter prior to said step (2) and subjecting the filtrate obtained in step (2) to sterile filtration.

10. (Previously Presented) The method according to claim 1, further comprising sterile filtering said solution obtained in step (1).

11. (Previously Presented) The method according to claim 1, comprising using a virus removal filter capable of reducing the concentration of model viruses having a size of ca 20 to ca 40 nm with at lest 4 log during a spiking test.

12. (Original) Method of stabilizing pharmaceutical compositions of purified leukocyte α-interferon subjected to filtration on a virus removal filter, comprising using a polysorbate as a stabilizer.

13-15. (Cancelled)

16. (Previously Presented) The method according to claim 1, wherein said recovered filtrate contains said non-ionic detergent.

Claim 17. (Previously Presented) A method of removing and/or inactivating intact or non-intact bacteria, viral material, or prions from a pharmaceutical composition of a purified biologically active interferon protein comprising:

(1) adding to a solution of the purified interferon protein a non-ionic detergent comprising polysorbate in an amount of 0.05 to 1 g/l;

(2) subjecting the solution containing the non-ionic detergent to filtration on a virus removal filter with a pore size of 10 to 40 nm; and

(3) recovering the filtrate.

18. (Previously Presented) The method according to claim 17, wherein non-enveloped viruses, and/or prions are removed and/or inactivated.

19. (Previously Presented) The method according to claim 1, wherein during the filtration on a virus removal filter there is no plugging of the filter.

20. (New) The method according to claim 1, wherein said non-ionic detergent comprises polysorbate in an amount of 0.1 to 0.5 g/l.

21. (New) The method according to claim 17, wherein said non-ionic detergent comprises polysorbate in an amount of 0.1 to 0.5 g/l.

REMARKS

Claims 1-3, 5-12 and 16-21 are pending. No new matter has been added by way of the present amendment. For instance, the subject matter amended to claims 1 and 17 is supported by originally filed claim 4. Claims 2 and 3 have been amended to parallel amendments made to claim 1. New claims 20 and 21 are supported by the present specification at page 7, lines 8-12 and page 8, lines 18-19. Lastly, claims 4 and 13-15 have been cancelled. Accordingly, no new matter has been added.

In view of the following remarks Applicants respectfully request that the Examiner withdraw all rejections and allow the currently pending claims.

Issues under 35 USC §§ 102(a) and 102(e)

The Examiner has rejected claims 1-4, 11, 16 and 19 under 35 USC § 102(a) and 102(e) as being anticipated by Van Holten et al., U.S. Patent No. 6,096,872 (hereinafter referred to as Van Holten '872). Applicants traverse this rejection.

Van Holten '872 discloses that a non-ionic excipient (corresponding to the non-ionic detergent of the present invention) is used as a processing aid during viral reduction or viral clearance of large biomolecules. The advantages of the processing aids include:

> (1) processing time is greatly reduced and concomitantly yield is greater, since the processing aids shift the equilibrium away from protein dimer,

> trimer and aggregate formation, which allows the product to be processed at an increased protein concentration, (2) the ability to use smaller pore size membrane allowing for greater assurance of viral clearance of the smaller non-enveloped virus, (3) the immunoglobulin processed through the membrane is not altered in IgG subclass or stability, and (4) processing equipment and therefore manufacturing floor space can be optimized for highest product yield per filter area.

(column 5, lines 27-38 of Van Holten '872).

However, Van Holten '872 is very specific about the amount of excipient to be added. For instance, at column 6, lines 31-36, Van Holten '872 discloses:

> The non-ionic excipient of the instant invention (of Van Holten) may be present in the protein solution initially in the processing method in the range of from about **0.015 g/L to about 0.024 g/L**, most preferably about 0.02 g/L or 20 ppm. **Such concentrations of excipient during processing were determined not to affect viral clearance**. (emphasis added)

Thus, Van Holten '872 makes clear that the amount of non-ionic excipient should not exceed about 0.024 g/L. In fact, this amount of non-ionic excipient relates to the critical micellar concentration as explained at column 6, lines 36-43 of Van Holten '872:

> Particularly preferred of the non-ionic excipients is polysorbate 80, which is employed most preferably during processing at a concentration of 0.002% or 20 ppm for processing. This range is **at or within the critical micelle concentration** for polysorbate 80. In determining concentrations for other excipients in this invention, **guidance may be taken in the knowledge of the critical micelle concentration for that excipient**.

By this passage, Van Holten '872 makes clear that the amount of the excipient should not exceed the critical micellar concentration.

Applicants submit that all of the above discussion concerning Van Holten '872 teaches away from the present invention. The present claims require that the non-ionic detergent comprise a polysorbate in an amount of 0.5 to 1 g/l. The minimum amount (0.05 g/l) of this range is twice the maximum amount of 0.025 g/l disclosed as permissible by Van Holten '872.

Normally, and according to Van Holten '872, one would keep the amount of non-ionic detergent below the critical micellar concentration, because above such concentration, the non-ionic detergent form micelles of varying sizes, which penetrate very slowly (present specification, page 4, lines 6-8). However, according to the present invention, the non-ionic detergents are added to pharmaceutical compositions in concentrations above the critical micellar concentration.

In summary, the present claims are distinguished from Van Holten '872. Reconsideration and withdrawal of this rejection are respectfully requested.

Issues under 35 USC § 102(b)

The Examiner rejected claims 13-15 under 35 USC § 102(b) as being anticipated by Yuen et al., WO 96/11018. Applicants traverse this rejection and submit that claims 13-15 have been

cancelled. Thus, this rejection is moot. Reconsideration and withdrawal thereof are respectfully requested.

The Examiner has also rejected claims 13-15 under 35 USC § 102(b) as being anticipated by Hiratani et al., U.S. Patent No. 5,173,415 (hereinafter referred to as Hiratani '415). Applicants traverse this rejection and submit that claims 13-15 have been cancelled. Thus, this rejection is moot. Reconsideration and withdrawal thereof are respectfully requested.

Issues Under 35 U.S.C. §103(a)

The Examiner has rejected claims 9 and 10 under 35 USC § 103(a) as being obvious over Van Holten '872 in view of the "Whatman Labsales Catalog, 1992". Applicants traverse this rejection.

As discussed above, Van Holten '872 fails to disclose using a non-ionic detergent comprising polysorbate in an amount of 0.5 to 1 g/l. In fact, Van Holten '872 makes clear that the amount of non-ionic detergent (excipient of Van Holten '872) should not exceed about 0.024 g/L. Thus, there is no motivation to utilize the presently claimed amounts of non-ionic detergent. The secondary reference of "Whatman Labsales Catalog" fails to provide motivation to increase the amounts of non-ionic detergents into the presently claimed range and in fact, any such suggestion would be ignored in view of the disclosure of Van Holten '872.

Accordingly, no *prima facie* case of obviousness exists. Reconsideration and withdrawal of this rejection are requested.

The Examiner has also rejected claims 1-8 and 11-19 under 35 USC § 103(a) as being obvious over Hiratani '415. Applicants respectfully traverse this rejection.

In the Office Action dated December 30, 2002, Applicants submit that the Examiner has erred in the review of the previously submitted Declaration. The Examiner notes and acknowledges that the differences observed by the Applicant are statistically significant and of industrial importance. Nevertheless, the Examiner still asserts that these results are not "unexpected" based on the results in Table 5 of Hiratani '415. Applicants disagree.

A close inspection of Table 5 of Hiratani '415 reveals that the Examiner has apparently looked at the wrong lines. The Examiner suggests that Hiratani '415 would teach that the recovery rate for a filter pretreated with albumin would be 53%. That value is given on the third line of the Table and it was obtained for a 0.5% solution of dextran, not for albumin. For a 0.5% solution of human serum albumin the recovery rate was, in fact, 96.3%. That is, it was even better for albumin than for a 0.5% solution of the polysorbate Tween 80 (91.4%). This means that, upon closer examination, the Hiratani '415 reference teaches away from the present invention. That is, one of ordinary skill in the

art would have expected albumin to work at least as well as a polysorbate.

However, as the experimental data of the present invention reveals, albumin will, in solution with biologically active proteins, significantly reduce the yield of those proteins when the solution is filtered on a nanofilter (please refer to Table 1 on page 13 of the present specification). This is due to the fact that albumin will cause clogging of the filter as discussed in the last paragraph of page 13 and as illustrated in Figure 3 of the present application.

Thus, Hiratani '415 claims that albumin and detergents are equally effective in improving yield of biologically active proteins when they are used for coating of filters before filtration. In contrast, in the present invention, it has been discovered that the yield of biologically active proteins was statistically higher when detergent was added to the solution before filtration as compared to the addition of albumin.

In summary, there exists no *prima facie* case of obviousness based upon Hiratani '415. However, even if the Examiner has hypothetically established a *prima facie* case of obviousness, the unexpected results of the present invention compared to Hiratani '415 rebut any hypothetical *prima facie* case of obviousness. Reconsideration and withdrawal of this rejection are requested.

The Examiner has also rejected claims 9 and 10 under 35 USC § 103(a) as being obvious over Hiratani '415 in view of the "Whatman

Labsales Catalog, 1992". Applicants respectfully traverse this rejection.

As indicated above, distinguishing features exist between the present claims and Hiratani '415. The secondary reference of "Whatman Labsales Catalog" fails to cure these deficiencies. Moreover, the unexpected results of the present invention compared to Hiratani '415 is unaffected by the secondary reference. Accordingly, this rejection is moot. Reconsideration and withdrawal thereof are requested.

The Examiner has rejected claims 1-19 stand rejected under 35 USC § 103(a) as being obvious over Georgiades, U.S. Patent No. 4,732,683 (hereinafter referred to as Georgiades '683) in view of Manabe, U.S. Patent No. 4,808,315 (hereinafter referred to as Manabe '315). Applicants respectfully traverse this rejection.

Applicants believe that the Examiner has misinterpreted Applicants previous arguments. It is a misunderstanding that Applicants have stated that detergents disclosed in Georgiades '683 would be effective only against Sendai virus. It is generally known that detergents inactivate all kinds of enveloped viruses, that is, viruses that carry a lipid envelop. The efficiency of detergents is based on the dissolution of the lipid envelop of the virus. In contrast, detergents do not inactivate non-enveloped viruses, that is viruses that do not carry a lipid envelop. Such detergent-resistant viruses include, for example, parvoviruses and hepatitis A virus.

Applicants wish to emphasize that the present invention is not based upon virus inactivation by detergents but rather removal of viruses by viral filtration in the presence of a non-ionic detergent comprising a specific amount of polysorbate. Thus, according to the present invention detergents and filtration are not directed to the same end. Rather, detergents do not have a primary role in virus removal, rather, they unexpectedly render the filtration of biologically active proteins through virus removal filters much more efficient. That is, addition of detergents to the solution prior to virus removal filtration prevents filter clogging and improves protein recovery. Neither Georgiades '683 nor Manabe '315 suggests or disclose that detergents are able to prevent protein losses or clogging of the filter. Thus, neither of the cited references whether taken alone or in combination, suggest or disclose this feature of the present invention. Accordingly, there exists no *prima facie* case of obviousness.

Moreover, due to the differences between the present invention and Georgiades '683, the present invention achieves unexpectedly superior results as illustrated in the Declaration under 37 CFR 1.132 (dated October 15, 2002) submitted on October 17, 2002. A review of the Declaration reveals that the difference in recovery between detergent compared to albumin was statistically significantly higher in the presence of detergents rather than albumin. In Table 1 of the present application only

one pair of experiments was carried out. However, when all experiments carried out are analyzed, protein recovery was significantly higher in the presence of detergent than in the presence of albumin ($p<0.05$, t test). This data has been presented in the attached Declaration.

Accordingly, this rejection is moot. Reconsideration and withdrawal thereof are requested.

In view of the above, Applicants respectfully submit that the presently pending claims define allowable subject matter. Accordingly, the Examiner is respectfully requested to withdraw all rejections and allow the currently pending claims.

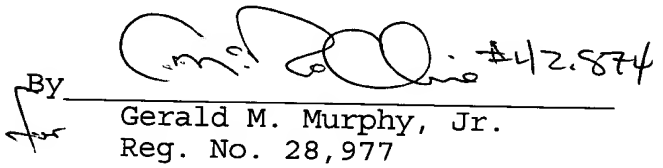
If the Examiner has any questions or comments, please contact Craig A. McRobbie, Registration No. 42,874 at the offices of Birch, Stewart, Kolasch & Birch, LLP.

If necessary, the Commissioner is hereby authorized in this, concurrent, and future replies, to charge payment or credit any overpayment to Deposit Account No. 02-2448 for any additional

fees required under 37 C.F.R. § 1.16 or under § 1.17; particularly, extension of time fees.

Respectfully submitted,

BIRCH, STEWART, KOLASCH & BIRCH, LLP



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